

**Amendments to the Claims:**

This listing of claims will replace all prior versions, and listing, of claims in the application:

**Listing of Claims:**

1. (Currently amended): A method of preparing a composition having reduced bacterial virulence of a pathogenic bacteria, comprising:
- growing in culture medium a virulent bacteria having a DNA methyltransferase (Dam) activity;
- contacting the bacteria with an agent that prevents the bacteria's *dam* gene expression thereby altering the bacteria's native level of methylation of adenine in a GATC tetranucleotide of the bacteria, and thereby reducing virulence of the bacteria ; and
- separating the bacteria from said culture medium ~~and excess agent~~ and adding to it the separated bacteria a pharmaceutically acceptable excipient, wherein the bacteria are selected from the group consisting of:
- Bordetella,
- Haemophilus,
- Neisseria,
- Pasteurella,
- Shigella,
- Treponema, and
- Yersinia.
2. (Canceled)
3. (Previously presented): The method of claim 1, wherein the agent causes a deletion within the bacteria's *dam* gene.
4. (Canceled)

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5. (Canceled)
6. (Previously presented): The method of claim 1, wherein the agent causes an insertion within the *dam* gene.
7. (Canceled)
8. (Canceled)
9. (Previously presented): The method of claim 1, wherein the agent binds a native *dam* nucleic acid sequence of the bacteria and prevents expression of a Dam gene.
10. (Canceled)
11. (Previously presented): The method of claim 1, wherein the agent alters Dam activity of a pathogenic bacteria selected from the group consisting of *Neisseria meningitidis*, *Pasteurella multocida*, and *Shigella spp.*
- 12-15. (Canceled)
16. (Currently amended): The method of claim [[12]] 1, wherein the bacteria are *Y. pseudotuberculosis pseudotuberculosis*.
17. (Previously presented): The method of claim 1, wherein the agent alters native Dam activity of a pathogenic bacteria selected from the group consisting of *Shigella*, *Haemophilus*, *Bordetella*, *Neisseria*, *Pasteurella* and *Treponema*.
18. (Original): The method of claim 1, wherein the bacteria are *Haemophilus*.
- 19-23. (Canceled)

24. (Currently amended): A method of ~~treating~~ protecting against a pathogenic bacterial infection by inhibiting proliferation of the bacteria, comprising the steps of:

administering to a subject ~~infected with the pathogenic bacteria~~ a therapeutically effective amount of a vaccine composition comprising a pharmaceutically acceptable excipient and an active agent comprising an attenuated bacteria which is avirulent due to having therein an alteration in a *dam* gene that alters the bacteria's native level of DNA methyltransferase (Dam) activity[[.]] wherein both said pathogenic bacterial infection and said attenuated bacteria are from one member of the group consisting of:

*Bordetella*,

*Haemophilus*,

*Neisseria*,

*Pasteurella*,

*Shigella*,

*Treponema*,

*Yersinia*,

*Escherichia*,

*Vibrio*,

*Y. pseudotuberculosis*

and *Salmonella*.

25-27. (Canceled)

28. (Previously presented): The method of claim 24, wherein the agent comprises a *dam* genetic construct that expresses Dam activity thereby increasing methylation of adenine in a GATC tetranucleotide in the bacteria, thereby ~~inhibiting~~ proliferation of the bacteria.

29. (Original): The method of claim 24, wherein the subject is a mammal.

30. (Original): The method of claim 24, wherein the subject is a human.

31. (Original): The method of claim 24, wherein the administering is by a route selected from the group consisting of oral, injection, inhalation and topical.

32. (Currently amended): The method of ~~treating~~ protecting against bacterial infection in an individual comprising administering to the individual a vaccine for producing an immune response to an immunogenic attenuated bacteria having a deletion in its DNA methyltransferase gene thereby reducing methylation of adenine in a GATC tetranucleotide in the bacteria, thereby inhibiting the virulence of the bacteria, said vaccine comprising said immunogenic attenuated bacteria and a pharmaceutically acceptable excipient, wherein both said pathogenic bacterial infection and said attenuated bacteria are from one member of the group consisting of:

Bordetella,

Haemophilus,

Neisseria,

Pasteurella,

Shigella,

Treponema,

Y. pseudotuberculosis,

Escherichia,

Vibrio,

Yersenia

and Salmonella.

33-46. (Canceled)

47. (Previously presented): The method of claim 1 wherein the agent is a polynucleotide.

48. (Currently amended): A method of ~~treating~~ protecting against a pathogenic bacterial infection by inhibiting proliferation of the bacteria, comprising the steps of:  
administering to a subject ~~infected with the pathogenic bacteria~~ a therapeutically

effective amount of a vaccine preparation comprising a pharmaceutically acceptable excipient and an active agent comprising a bacteria having therein an alteration in its native *dam* gene that alters the bacteria's native level of DNA methyltransferase (Dam) activity, said alteration selected from the group consisting of: an insertion in the *dam* gene[[:]] , a deletion in the *dam* gene[[:]] , and an additional copy of a *dam* gene for overproducing Dam methylase wherein both said pathogenic bacteria and said attenuated bacteria are from one member of the group consisting of:

*Bordetella*,

*Haemophilus*,

*Neisseria*,

*Pasteurella*,

*Shigella*,

*Treponema*,

*Yersinia*,

*Escherichia*,

*Vibrio*,

*Y. pseudotuberculosis*

and *Salmonella*.

49. (Previously presented): The method of claim 47 wherein said bacteria having an alteration is selected from the group consisting of: *Escherichia*, *Vibrio*, *Yersinia* and *Salmonella*.

50. (Previously presented): The method of claim 48 wherein said bacteria having an alteration is of a different species than said pathogenic bacterial infection.

51. (Currently amended): A method of preparing a composition having reduced bacterial virulence of a pathogenic bacteria, comprising:  
growing in culture medium a virulent bacteria having a DNA methyltransferase (Dam) activity;  
contacting the bacteria with an agent that increases the bacteria's *dam* gene

expression thereby altering the bacteria's native level of methylation of adenine in a GATC tetranucleotide of the bacteria, and thereby reducing virulence of the bacteria; and separating the bacteria from said culture medium and ~~any excess agent therein~~ and adding to ~~[[it]]~~ the bacteria a pharmaceutically acceptable excipient wherein the bacteria are selected from the group consisting of:

*Bordetella,*

*Haemophilus,*

*Neisseria,*

*Pasteurella,*

*Shigella,*

*Treponema, and*

*Y. pseudotuberculosis.*

52. (New): The method of claim 24, wherein the bacteria are selected from the group consisting of:

*Bordetella,*

*Haemophilus,*

*Neisseria,*

*Pasteurella,*

*Shigella,*

*Treponema, and*

*Y. pseudotuberculosis.*

53. (New): The method of claim 32 wherein the bacteria are selected from the group consisting of:

*Bordetella,*

*Haemophilus,*

*Neisseria,*

*Pasteurella,*

*Shigella,*

*Treponema, and*

*Y. pseudotuberculosis*.

54. (New): The method of claim 48 wherein the bacteria are selected from the group consisting of:

*Bordetella*,

*Haemophilus*,

*Neisseria*,

*Pasteurella*,

*Shigella*,

*Treponema*, and

*Y. pseudotuberculosis*.

Amended  
at 12/05  
HJ